Asian Journal of Plant Sciences

ISSN 1682-3974

Asian Journal of Plant Sciences
Volume 1 Number 3: 283-287, 2002

Palynological Study of the Genus *Tragopogon* from Pakistan

Sohail Jamil Qureshi, Abdul Ghani Awan, Mir Ajab Khan and Sofia Bano
Department of Biological sciences, Quaid-I-Azam University Islamabad, Pakistan

Abstract: Pollen morphology of two different species, *Tragopogon dubius* and *Tragopogon gracilis*, belonging to genus *Tragopogon* of family Asteraceae was studied from Pakistan. Morphology of pollen grains of each of the species is based on specimens selected at random. Proposed characters i.e. grain, shape of pollen grain, equatorial view, polar view, equatorial diameter (E), polar diameter (P), P / E ratio, length of colpus, exine surface, exine thickness, inter poral distance, inter spinal distance, inter spinal outline, length of spines, number of spines between colpi in each species were recorded for comparison. At species level, micromorphological differences and distribution of surface pattern, shape and size of pollen have been found to exist. The pollen grains are consistently echinate, trizonocolporate but in *Tragopogon dubius* pollen grains are tetrazonocolporate. Maximum, equatorial view, polar view, equatorial diameter (E), polar diameter (P), P/ E ratio, length of colpus, exine surface, exine thickness, inter poral distance, inter spinal distance, inter spinal outline was observed in and *Tragopogon gracilis* and length of spines is maximum in *Tragopogon dubius*. This study demonstrates the potential of pollen studies in distinguishing some taxonomic groups in the Asteraceae.

Key words: Palynology, *Tragopogon*, pollen grain, Asteraceae

Introduction

Pollen morphology of the *Lactuceae* (Cichorieae) is probably the more distinctive tribe in the family Asteraceae. The ligulate corolla, milky sap and echinolophate pollen form a unique combination of characters which it can be readily distinguished from the rest. This tribe consists of about 70 genera and 2300 species (Tomb, 1977). Stebbins (1953) proposed a natural system of classification for this tribe using geographical distribution, pollen morphology and chromosomal data in addition to traditional morphological characters. This method produced eight subtribes (I) Scolyminae, (ii) Cichorinae, (iii) Microseridinae, (iv) Stephanomeriinae, (v) Dendroseiidinae, (vi) Scorzonerinae, (vii) Leontodontinae and (viii) Crepidinae. Jeffrey (1966) revised Stebbin's classification recognizing the importance of microcharacters like length of collector hairs on the style, shapes of hairs on stigmatic surfaces and pubescence on the corolla tube. He divided this tribe into five groups, eleven subgroups, and 23 series.

Many workers regard pollen grains of Lactuceae as "Liguliflorae-type" (Faegri and Iverson, 1975; Moore and Webb, 1978) and contrast this with a "Tubiliflorae-type" for most of the reminder of the family Asteraceae. Wodehouse (1928,1935) examined a large number of taxa of this tribe primarily in an effort to formulate phylogenetic trends within the tribe. His studies led to the characterization of several basic echinolophate patterns common in the Lactuceae. Pausinger (1951) divided the tribe into two main types based on pollen characters. His Leontodon type was characterized by the possession of poral lacunae, and this *Tragopogon* type of the lack of poral lacunae and the aboral ones communicating to form long lacunae. While working on the comparative pollen morphology of *Sonchus*, Boulos (1960) found that this genus was closely related to *Launaea*. Tomb *et al.* (1974) studied the pollen morphology of Stephanomeriinae and showed that pollen grains of most of the tribe were echinolophate or tricolporate with same, or almost the same, number and shape of lacunae and demonstrated strikingly different exine stratification in several genera. Feuer (1974) examined the pollen grains of Microseridinae, which, in contrast with the stephanomeriinae, were predominantly echinolophate. Skvarla *et al.* (1977) summarized much that was taken about pollen structure in the Asteraceae, where two major pollen types were categorized, namely, anthemoid and helianthoid, with various subtypes. Taxonomic, evolutionary and functional studies of the Asteraceae pollen grains on the basis of ultrastructure and sculpture were made by Bolick (1978), who noted two basic exine patterns: The caveate helianthoid and non-caveate Anthemoid. El Ghazaly (1980) studied the pollen grains of 35 species of the subtribe Hypochoeridinae. Regarding the sub tribe Scorzonerinae with reference to its taxonomic significance, Blackmore (1982) recorded seven pollen types, which could be distinguished by a key constructed on the basis of the number and arrangement of the lacunae of the grains. Blackmore (1984) further dealt with pollen morphology of a large number of taxa of the tribe Lactuceae and recognized seven distinct pollen types, which were further subdivided into smaller groups on the basis of distinguishing characters. According to Clark *et al.* (1980) pollen grains of the *Astereae* have been characterized as basically helianthoid, spherical or slightly flattened, tricolporate, and uniformly echinate, having internal foramina, with varying proportions of abnormalities in size and colpus number (Wodehouse, 1930, 1935; Skvarla and Turner, 1966; Skvarla *et al.*, 1977). Pollen of the few genera examined to date has been difficult or impossible to distinguish by light or electron microscopy (Skvarla *et al.*, 1977). However, in conjunction with systematic studies of *Haplopappus* and related genera in the subtribe Solidaginae, we have found a few cases of significant variation in pollen size, spine length, and the number of spine rows between colpi. These characters indicate a potential for utilizing pollen characters in at least some systematic studies in the Astereae.

According to Larson and Lewis (1961), interest in pollen morphology has increased as its useful application in systematics, paleoecology, paleobotany and inhalent allergy has been increasingly recognized. Pollen morphologists have responded to the need, created by this widespread application, for a more critical comparative analysis of pollen wall structure, and for an expansion in the number of recognized systematically and phylogenatically significant wall characteristics. In this response, successful use has been made of phase and ultraviolet microscopy in addition to more sophisticated light microscopy. The reader is directed to the excellent review of pollen wall analysis in Wodehouse (1935) to Erdtman (1959) for a demonstration of the results of ultraviolet microscopy and to Stix (1960), whose study of pollen walls in the Composite is of real systematic value.

Pollen morphology has also gained much from electron-microscopic studies of sectioned pollen grains. The division of the pollen wall into ektexine, endexine and intine, as described in Faegri and Iversen (1950) (a basic discussion of the pollen wall is found in this text and in Erdtman [1952]), was observed in the pioneer electron microscopic studies of Muhlethaler (1953). However, the sections in these studies were too thick to reveal fine structural detail. During the initial period of rapid improvement of fixing, embedding and sectioning techniques in the field of electron microscopy, the electron micrographs published by Afzelius (1954, 1955, 1956) and Afzelius *et al.* (1954) were made from sections of sufficient thinness to reveal fine structural detail and differentiation of wall layers.

Pollen characters were used by Stebbins (1953) and monographic treatments since than have used them to some extent. Wodehouse (1935) characterized the basic exomorphology of *Lactuceae* pollen and described two basic pollen types (echinate and lophate). There have been several careful light-microscopic studies of pollen since the wodehouse study (i.e., Pausinger, 1951; Saad, 1961). However, there has not been an in-depth study of the tribe using modern electron microscopy techniques (SEM and TEM) until recently (Tomb *et al.*, 1974; Feuer and Tomb, in prep. Subtribes Stephanomeriinae and microseridinae, respectively). These studies have shown that pollen of most of the tribe is echinolophate or tricolporate, with the same or almost the same numbers and shapes of lacunae, and demonstrated strikingly different exine stratification patterns in several genera. Pollen characters have been careful at several levels in the systematics of the tribe. For example, in the Stephanomeriinae aperture-shape supports the division of the subtribe into two phyletic lines, a division suggested by Stebbins (1953). In the ditypic genus Glyptopleura (Stephanomeriinae), pollen morphology (internal and external) of the two species is quite different (Tomb *et al.*, 1974). At the population level, using herbarium material, pollen characters have been used to map hybrids and polyploids (Tomb, 1970 and Northington, 1971). Hybrids (as well as apomictic plants) usually produce a high frequency of aborted and irregular grains. Polyploids generally have larger pollen and are often tetracolpate. Among the important contributions of these studies were the observations of variation in the fine structure of the ektexine, the size of the wall substance particles (50-60 A), and the stratified nature of the exine. In both gymnosperms and angiosperms plants, an inner layer of the exinewas observed to be laminated and less compact than the other layers. This layer was interrupted as being a part of the endexine (homologous to the endonexine (Erdtman, 1952; 1960) and as having phylogenatic value. In the plants studied, pollen grains of the gymnosperms species were found to have a laminated layer considerably greater in thickness than that of any angiosperms plants studied. From this observation, a reduced laminated layer was considered phylogenetically advanced. In his electron microscopic study of the pollen wall in *Saintpaulia ionantha*, Ehrlich (1958) presented further evidence of a laminated layer between the intine and ektexine and also observed that this laminated layer made up the aperture membrane. In Ehrlich's study, this layer is interpreted as an independent wall layer, the mesine. Some pollen morphologists now recognize the pollen wall to be composed of ektexine, endexine, mesine and intine. The potential systematic value of the mesine or an analogous layer is made obvious as Rowley (1959) found no evidence of a mesine in the pollen wall in the Commelinaceae. The possibility that the lack of a mesine resulted from the techniques employed by Rowley (formalin fixation, OsO_4 staining and methacrylate embedding) is ruled out as they were the same as those used by Ehrlich. Rowley also reported observing a mesine in pollen of dicotyledonous plants.

The fine structure and wall stratification of fossilized polen from the Eocene has also been analyzed by electron microscopy. Ehrlich and Halls's (1959) exciting study shows that fossilized pollen walls retain structural details, and indicates that the evolutionary development of the pollen wall may be subject to direct study. Recognizing the potential systematic and phylogenatic value of the pollen wall, especially the layer presently recognized as the mesine, a comparative study of *Parkinsonia aculeata* pollen wall was initiated. This species was chosen because, in an examination of the fine structure of the cytoplasm of this pollen, a mesine-like layer and an aperture membrane, more complex in fine structure than any previously reported, were observed.

Gramineae is one of the largest family of flowering plants, comprising of 620 genera and c. 10000 species is widely dispersed in all the regions of the world (Willis, 1973). In Pakistan, it is represented by 158 genera and 492 species (Stewart, 1972; Cope, 1982). Pollen grains of Gramineae also do not show much deviation from this contention and has long been recognized as remarkably uniform. Therefore palynology plays a little role in the taxonomy of this family (Wodehouse, 1935; Rowley, 1960). However according to Wodehouse (1935) two characters viz., grain size and sexine pattern use of some significance. Firbas (1937) used the grain sizeas a basic character to separate wild and cultivated grasses. Rowley (1960) while studying the fine structure of some of the grass pollen used the arrangement of the spinules on the ektexine as a key character for delimiting various taxa.

Faegri and Iversen (1964) in their study of grass pollen, found two different types of sexine i.e. scabrate and areolate. Anderson and Bertelsen (1972) and Grant (1972) also used these types to distinguish various members of the tribe Triticeae, *Zea and Tripsacum,* respectively. Page (1978) in his scanning electron microscopic survey of grass pollen further divided these two basic types on the basis of the proximity of granules, whether they are closely or widely spaced, while the fused type are differentiated on the basis of height of granules. Chaturvedi (1971) has reported 4 types of grains in *Saccharum robustum* viz., (I) normal monoporate grains (ii) double grains with two pores on either side of the dumbbell shaped pollen grains (iii) single diporate grains and (iv) double grains with single pore.

According to Memon (1984), the Scanning Electron Microscope has revealed micro-morphological details of surface patterns of pollen grains, which are not resolved either by the transmission electron microscope or by an optical microscope, the sectional study of the exine can not be as accurately studied by the scanning electron microscope as by the transmission electron microscope. The reveal advantage of the scanning electron microscope in the sectional study of exine in the sectional study of exine lies in the realm of middle magnitude structures in three dimensions. However, the optical microscope is very useful for basic study of pollen morphology, especially for studying the details of aperture. It was noted that the surface pattern of pollen grains was more completely analyzed by the scanning electron microscope than the optical microscope which exaggerates some features or does not reach the full depth of focus of exine sculpture. The remarkable architecture of pollen exine is known to be distinctive for different texa and each species retains its specific statistic which can be characterized on the basis of pollen morphology (Memon, 1985). Erdtman (1963) segreted two South American genera *Abolboda* and *Orectanthe* from the family Xyridaceae and referred them to a special family Abolbodaceae solely on the basis of pollen morphological characters. The pollen morphology of family Proteaceae has revealed a wide spread heterogeneity and many genera including nearly all those with large number of species often lack unique combinations of pollen characters that could distinguish them from other genera of the family. Although some tribes and sub tribes with a small number of genera show homogeneity in their pollen morphology, nonetheless, they could not be separated from one another because invariable overlapping of surface pattern, shape and size of pollen grains was present throughout the family (Memon, 1984).

Zahur *et al.* (1975-78) had provided a commendable quantity of basic and applied information in this line by describing the pollen grains of 486 angiosperm species. They also described the size range of pollen grains of Gramineae. Erdtman (1961) found uniporate pollen grains of *Hordeum vulgare* with 3 μ diameter. Ashraf (1973) studied some medicinal plants palynologically. Wodehouse (1965) reported pollen grains of *Avena barbata* as an important cause of hay fever. Malik *et al.* (1964) studied pollen morphology of seventy-five Pakistani medicinal plants. They observed that the pollen grains are of various shapes i.e. varying from spheroidal to prolate with polar or lateral germinal exits. They also reported different measurements of pollens. Elisens (1986) studied morphological variation among 12 new world genera in tribe Antirrhineae (Scrophulariaceae) with light and scanning electron microscopy. Pollens from 29 American species have mean polar diameter that range from 17 to 26 μm, have a tectate structure with perforate micro reticulate or reticulate surface pattern and are subspheroidal to proloate and trizono-colporate with fusiform or narrowly oblong, colpi that are free or occasionally fused at the poles.

The pollen characteristics of some medicinal plants was made by Malik *et al.* (1964). This seems to be first palynological contribution from Pakistan. Although Erdtman (1952) has very elaborately covered this neglected field yet regional pollen flora remained unexplored. Khan and Bhutta (1965) studied the pollen grains of honey. In a review paper Bhutta(1968) emphasized the need of this important field of science. Soon after Khan and Memon (1970) gave an account of the pollen morphology of certain Leguminous plants of Jamshoro, Sind. Taking into account the applied form of Palynology it was found desirable to produce series of papers dealing with the fundamental Palynology of the plants growing in the Punjab. The fundamental palynology will certainly help the applied aspects.

According to Ali (1988), in most of the plant groups in Angiosperms, at maturity, all the pollen grains are free from each other. As the prime function of the pollen grains is to provide the male gametes to the female counter part, in order to facilitate fertilization and ultimately the formation of the seed, such as category seems logical. However, it is also well known that in some plant groups, the pollen grains do not separate at maturity and remain associated together. The term pollen aggregate is used for all types of compound pollen grains and for various types of situations where pollen grains are dispersed in-groups. Char *et al.* (1973) reviewed the evolutionary trends in pollen organization and discussed their adaptive significance. In the present investigation, the pollen morphology of the genus *Crepis*, all Pakistan, of the tribe Lactuceae, have been studied systematically. The present writers decided to contribute as much as possible and to start with different species of the genus Asteraceae. The present study was undertaken to evaluate the morphological differences in the pollen grains of family Asteraceae.

Materials and Methods

Pollen morphology: Pollen morphology of six taxa of Tribe *Lactuceae* of Asteraceae (Composite) from Pakistan was studied. The florets from mature capitula were extracted, either from the herbarium specimens of Quaid-I-Azam University, Islamabad or fresh polliniferous material collected from the wild was used in this research during the year 2001.

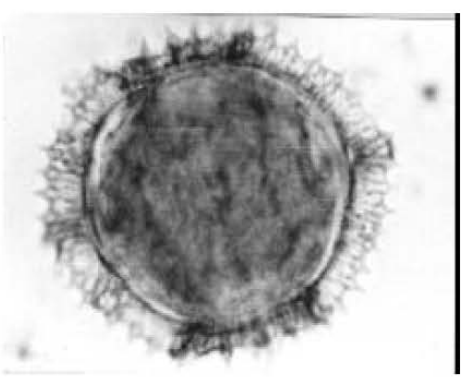
Pollen staining preparation: Pollen grains were stained with 1-% safranine mixed in glycerin jelly. Glycerin jelly was prepared by dissolving 70gm of gelatin in 42ml distilled warm water in a beaker. The beaker was placed in another metalloid pot, containing boiling water. The gelatin was stirred for 1-2 hours. After this operation, 35ml of gelatin was added in it, followed by 1gm of phenol crystals. This warm gelatin jelly was filtered using filter paper, 1-% safarinine solution was poured in it in 1:1 ratio. The homogenized mixture was preserved in a vial and was used for staining the pollen grains.

Pollen grain study by light microscopy: Florets taken from herbarium specimens were kept in distilled water in petri dishes for about 24 hours so as to soften them and were then used, while fresh material was used directly. The florets were separated from capitula and were placed in a few drops of distilled water on a clean glass slide. With the help of dissecting needles, the florets were opened, the extra material was removed and the anthers were opened, the extra material was removed and the anthers were crushed to release pollen grains on the slide. Anther wall material was discarded, while excess of water was removed with filter paper. Pollens were stained with 1% safranine mixed in glycerin jelly. The slide was placed on a hot plate and when the stain had completely melted, any bubble formed was carefully removed. Cover slip was placed on the prepared pollen glycerin jelly mixture. When cooled, the glass slide was labeled and the cover slip edges were sealed with white transparent nail varnish. The prepared slides were studied under the light microscope. Eight slides of each Taxon were prepared and complete set is kept in the Plant Taxonomy Lab, Department of Biological Sciences, Quaid-I-Azam University, Islamabad. Their photographs were taken with Nikon Apaphot Microscope (LM).
For the measurements of pollen grains, following characters were noted:- grain, shape of pollen grain, equatorial view, polar view, dimensions, equatorial diameter (E), polar diameter (P), P / E ratio, length of colpus, exine surface, exine thickness, inter poral distance, inter spinal distance, inter spinal outline, length of spines, number of spines between colpi,

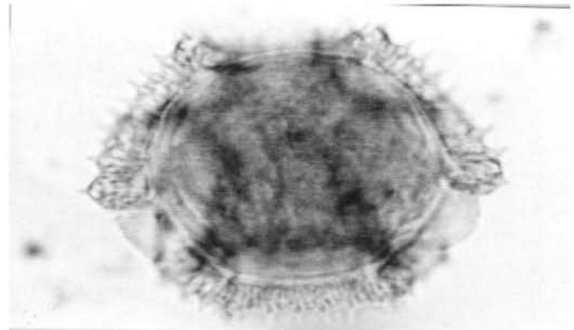
Results

Tragopogon gracilis D. Don. (Plate 1)
Grain: Trizonocolpate
Shape of pollen grain: Spheroidal
In equatorial view: spheroidal
In polar view: Triangular
Dimentions:
Equatorial diameter (E): 45-(45.7)-46.4
Polar diameter (P): 42.5-(43.75)-45
P / E ratio: 0.95-(0.96)-0.97
Length of colpus: 5-(5.7)-6.4
Exine surface: Echinate or Spinate
Exine thickness: 0.5-(0.75)-1
Inter spinal distance: 0.5-(0.9)-1.3
Inter spinal outline: V shaped
Length of spines: 3.75-(4)-4.2
Number of spines between colpi: 8

Plate 1: Pollen light micrograph
Tragopogon gracilis D. Don.



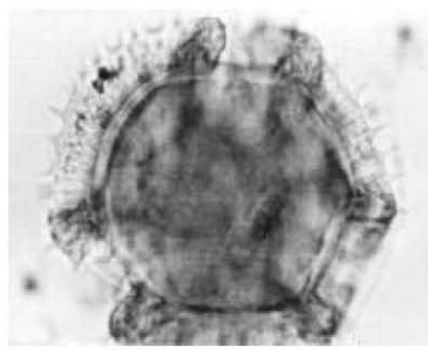
A : Equatorial view



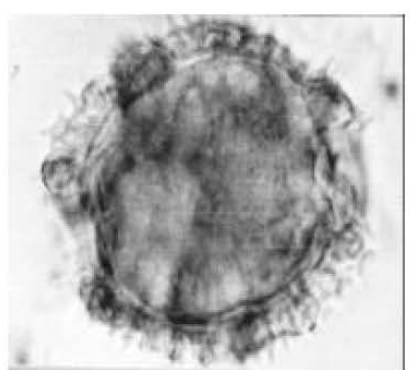
B: Polar view

Tragopogon dubius Scop. (Plate 11)
Grain: Tetrazonocolporate
Shape of pollen grain: Sub spheroidal
In equatorial view: Sub spheroidal
In polar view: Quadrangular
Dimentions:
Equatorial diameter (E): 58.5-(59.25)-60
Polar diameter (P): 54.4-(58.45)-62.5
P / E ratio: 0.93-(0.99)-1.04
Length of colpus: 8.75-(9.4)-10
Exine surface: Echinate or Spinate
Exine thickness: 3.75-(3.82)-3.9
Inter poral distance: 40-(43)-46
Inter spinal distance: 2.5-(3.1215)-3.75
Inter spinal outline: V shaped
Length of spines: 2.5-(2.75)-3
Number of spines between colpi: 10

Plate 11: Pollen light micrograph
Tragopogon dubius Scop.



A: Equatorial view



B: Polar view

Table 1: Minimum, maximum and average values of polar and equatorial diameter, P/E ratio and Colpi length of genus *Tragopogon*

Characters	Different Values	*Tragopogon dubius*	*Tragopogon gracilis*
Equatorial diameter	Minimum	58.5	45.0
	Maximum	60.0	46.4
	Average	59.25	45.7
Polar diameter	Minimum	54.4	42.5
	Maximum	62.5	45.0
	Average	58.45	43.75
P/E ratio	Minimum	0.93	0.95
	Maximum	1.04	0.97
	Average	0.98	0.96
Colpi length	Minimum	8.75	5.0
	Maximum	10.0	6.4
	Average	9.3	5.7

Table 2: Minimum, maximum and average values of inter polar diameter, inter spinal distance, length of spine and spine rows b/w colpi of genus *Tragopogon*

Characters	Different Values	*Tragopogon dubius*	*Tragopogon gracilis*
Exine thickness	Minimum	3.75	0.50
	Maximum	3.90	1.00
	Average	3.82	0.75
Inter poral distance	Minimum	40.00	25.00
	Maximum	46.00	28.00
	Average	43.00	26.50
Inter spinal distance	Minimum	2.50	0.50
	Maximum	3.75	1.30
	Average	3.12	0.90
Length of spines	Minimum	2.50	3.75
	Maximum	3.00	4.20
	Average	2.75	3.97
Number of spines b/w colpi		10.0	8.00

Discussion

In the present investigation the genus *Tragopogon* characteristic pollen grains were observed. In *Tragopogon gracilis* the pollen grains are trizonocolporate but in *Tragopogon dubius* the pollen grains are tetrazonocolporate. The pollen grain shape in equatorial view was spheroidal to subspheroidal, polar view is triangular in *Tragopogon gracilis* and quadrangular in *Tragopogon dubius*. Equatorial diameter varies from 45-58.5 μm (minimum) and 46.4-60 μm (maximum) (Table 1). The size of polar diameter varies between 42.5-54.4 μm (minimum) and 45-62.5 μm (maximum) (Table 1). The equatorial and polar diameter is minimum in *Tragopogon gracilis* 45 μm, 42.5 μm respectively. In *Tragopogon dubius* the equatorial diameter is 60 μm and polar diameter is 62.5 μm.

Colpus length is maximum in *Tragopogon dubius* 10 μm while least in *Tragopogon gracilis* 5 μm. In *Tragopogon* the pollen grains are echinate or spinate. In *Tragopogon dubius* the exine thickness maximum 3.9 μm while minimum in *Tragopogon gracilis* 0.5 μm (Table 2). In this research project, some more morphological parameters were considered for their application in plant taxonomy, which were found to be useful. Minimum value of interpolar distance were found in *Tragopogon gracilis* 25 μm but maximum value of interpolar distance were found in *Tragopogon dubius* 46 μm (Table 2). The maximum interspinal distance were found in *Tragopogon dubius* 3.75 μm while least in *Tragopogon gracilis* 0.5 μm. In *Tragopogon* the interspinal outline is V shaped. Spine length were found to be maximum in *Tragopogon gracilis* 4.2 μm but minimum in *Tragopogon dubius* 3 μm. Number of spine rows b/w colpi varies from 8-10 in different species of *Tragopogon* (Table 2)

Light microscopic observations could not clearly indicate the exine sculpturing. Bolick (1978) suggested that Scanning Electron Microscopic (SEM) studies should be carried out for obtaining many characters of great taxonomic importance. Not only the general morphology but also pollen morphology is of significance in species delimitation and pollen characters are correlated with morphological features (Stix, 1960). Palynology can play a very important role in solving the taxonomic problems if the pollen characters are co-related with morphological characters to become a qualitative character.

It is concluded that pollen morphology can not be solely used as the base of taxonomic classification of the family. However, if it is accepted that pollen morphology shows evolutionary sequences comparable to those in other organs, then it may need to be given as much weight as any other morphological character. On this assumption, in the pollen morphology of the species examined, some inconsistencies and alternative relationships have been suggested in correlation with the recent taxonomic classification proposed by Johnson and Briggs (1975). The general features of the Asteraceae, taken together are not repeated in other families. This gives Asteraceae (Compositae) a unique taxonomic status. To quote "Combined efforts of systematic, evolutionary and ecological studies are still necessary to help us understand the evolution of this fascinating family. Lactuceae is a tribe of Asteraceae, some plants in this tribe are of medicinal importance. Palynological studies of Lactuceae from Pakistan are carried out for the first time. The study of pollen morphology has assumed great significance in plant taxonomy and the advancements in microscopy have led to the effective use of new pollen morphological parameters for taxonomic purposes. This research project was conducted to examine the value of pollen morphology in the taxonomy of Lactuceae and if the palynological characters are correlated to the morphological features, then they have great significance in taxonomy and may be considered as base for taxonomic decisions. For structure and pattern describing different characters, terminology followed is that of Erdtman (1969) and Nair and Lawrance (1985). For clear understanding to the taxonomic status of the species following palynological characters like grain class, shape in equatorial and polar view, equatorial and polar diameter, P/E ratio, colpus length, exine surface, exine thickness, interporal distance, interspinal distance, interspinal outline, length of spine, number of spines b/w colpi were also considered. It is hoped that added information of pollen grains will help in taxonomic studies of *Tragopogon*. Among the members of Lactuceae the general pollen morphology is similar i.e. the exine surface is echinate (spinate) or echinate (spinulate), the pollen grain is trizonocolporate but in some species tetrazonocolporate.

References

Afzelius, B. M., 1954. La structure du sporoderme vue au microscope electronique. Rapports et communications. Section, 6: 241. 8 Congres International de Botanique.

Afzelius, B. M., 1955. On the fine structure of the pollen wall in Clivia miniata. Bot. Notiser., 108: 138-145.

Afzelius, B. M., 1956. Electron microscope investigations into exine stratification. Grana Palynologica, 12 : 22-37.

Afzelius, B. M., G. Erdtman and F. S. Sjostrand, 1954. On the fine structure of the outer part of the spore wall of Lycopodium clavatum as revealed by the electron microscope. Svensk Bot. Tidskr., 48: 155-161.

Ali, S. A., 1988. The Functional Significance of Pollen Aggregates in Angiosperms. Pak. J. Bot., 20: 21-44.

Anderson, S. Th. and F. Bertelsen, 1972. Scanning electron microscopic studies of cereals and other grasses. Grana, 12: 79-86.

Ashraf, M., 1973. Pollen morphology of certain medicinal plants of Punjab plains. M.Sc. Thesis, Pb. Univ., Lahore.

Bhutta, A. A., 1968. Palynology- the study of pollen and spores. Horizon, 1047-49.

Char, M. B. S., M. S. N. Swamy and M. C. Cheluviah, 1973. A critical evaluation of the adaptive significance of the monadsand compound grains in Angiosperms. J. Palynol., 9: 39-44.

Chaturvedi, M.,1971. Pollen grains of Saccharum robustum. Grana, 11: 87-90.

Clark, W. D., G. K. Brown and R. A. Mayes, 1980. Pollen morphology of haplopappus and related genera (Compositae- Astereae). Am. J. Bot., 67: 1391-13963.

Cope, T. A., 1982. Poaceae. In: E. Nasir and Ali, S. I. (Eds.) Flora of Pakistan, 143: 1-678, Islamabad.

Erdtman, G., 1969. Handbook of Palynology. An introduction to the study of pollen grains and spores. Munksgaar, Copenhagen.

Ehrlich, H. G., 1958. Electron microscope studies of *Saintpaulia ionantha* Wendl. Pollen walls. Exptl. Cell Res., 15: 463-467.

Ehrlich, H. G. and J. W. Hall, 1959. The ultrastructure of Eocene pollen. Grana Palynologica, 21: 32-35.

El Ghazaly, G., 1980. Palynology of Hypochoeridinae and Scolyminae (Compositae). Opera Bot., 58: 1-48.

Elisens, W. J., 1986. Pollen morphology and systematic relationship among new world species in tribe Antirrhineae (Scrophulariaceae). Am. J. Bot., 73: 1298-1311.

Erdtman, G., 1959. UV micrographs and photomicrographs from the palynological laboratory, Stockholm-Solna. Grana Palynologic, a21: 36-39.

Erdtman, G., 1961. Pollen and spore morphology plant taxonomy. *Pteridophyta,* pp: 272. Almqvist and Wiksell, Stockholm.

Erdtman, G., 1963. Palynology. In, Advances in Botanical Research. (Ed.) R.D. Preston, 1: 149-208.

Erdtman, G., 1952. Pollen morphology and plant taxonomy. Angiosperms. Chronica Botanica Co., Waltham, Mass.

Grant, C. A., 1972. A Scanning Electron Microscope Survey of Some Maydeae Pollen. Grana, 12: 177-184.

Jeffrey, C., 1966. Notes on Compositae 1. The Cichorieae in East Tropical Africa. Kew Bull., 18: 427-486.

Johnson, L. A. S. and B. G. Briggs, 1975. On the Proteaceae- the Evolution and Classification of a Southern Family. Bot. J. Linn. Soc., 70: 83-182.

Khan, K. M. and Butta, A. A., 1965. Pollen analysis of honey. All Pak. 17th Sci. Conf., Karachi.

Khan, K. M. and H. R. A. Memon, 1970. Palynological investigations of some members of family Leguminosae from Jamshoro (Sind), West Pakistan. Pak. J. Bot., 2: 25-35.

Larson, Donald A. and C. W. Lewis, 1961. Fine structure of Parkinsonia Aculeata Pollen. 1. The Pollen Wall in Am. J. Bot., 48: 934-943.

Muhlethaler, K., 1953. Untersuchungen iibr die Strukture der pollenmembran. Mikroskopie, 8: 103-110.

Nair, P. K. K. and R. Lawrernce, 1985. Pollen Morphology in Compositae. Kalyani Publishers. New Delhi. India.

Northington, D. K., 1971. Taxonomy of Pyrrhopappis a cytotaxonomic and chemosystematic study. Ph. D. Dissertation, University of Texas, Austrin.

Pausinger, F., 1951. Vom Blutenstaub der Wagwarten (die Pollengestaltung der Cichorieae). Carinthia, II 13: 3-47.

Page, J. S., 1978. The Scanning Electron Microscope Survey of Grass Pollen. Kew bulletin., 32: 313-319.

Saad, S. I., 1961. Pollen Morphology in The Genus Sonchus. Pollen et Spores, 3: 247-260.

Stix, E., 1960. Pollen morphologische Untersuchungen an Compositen. Grana Palynologica., 22: 41-114.

Tomb, A. S., D. A. Larson and J. J. Skvarla, 1974. Pollen morphology and detailed structure of family Compositae, tribe Cichorieae. 1 Subtribe Stephanomeriinae. Am. J. Bot., 61: 486-498.

Tomb, A. S., 1977. Lactuceae- systemativ Review. In: V.H. Heywood, J. B. Harborne and B. L. Turner (Editors), the Biology and Chemistry of Compositae., pp: 1067-1079. Academic press, London, New York.

Willis, J. C., 1973. A dictionary of the flowering plants and ferns. Cambridge Univ. Press., pp: 1245.

Wodehouse, R. P., 1930. Pollen grains in the identification and classification of plants-V. Haplopappus and other Astereae: the origin of their furrow configurations. Bull. Torrey Bot. Club, 57: 21-46.

Wodehouse, R. P., 1935. Pollen grains, their structure, identification and significance in medicine. Hafner Publishing company, New York, pp: 574.

Wodehouse, R. P., 1935. Pollen grains. McGraw-Hill, New York.

Wodehouse, R. P., 1965. Reprinted in 1965. Pollen grains New York. Hanfan and Inc. New York.

Wodehouse, R. P., 1928.The Phylogenetic value of pollen grain characters. Ann. Bot., 42: 891-834.